

SCHEMATIC REPRESENTATION OF DEN POLYPROTEIN MATURATION.

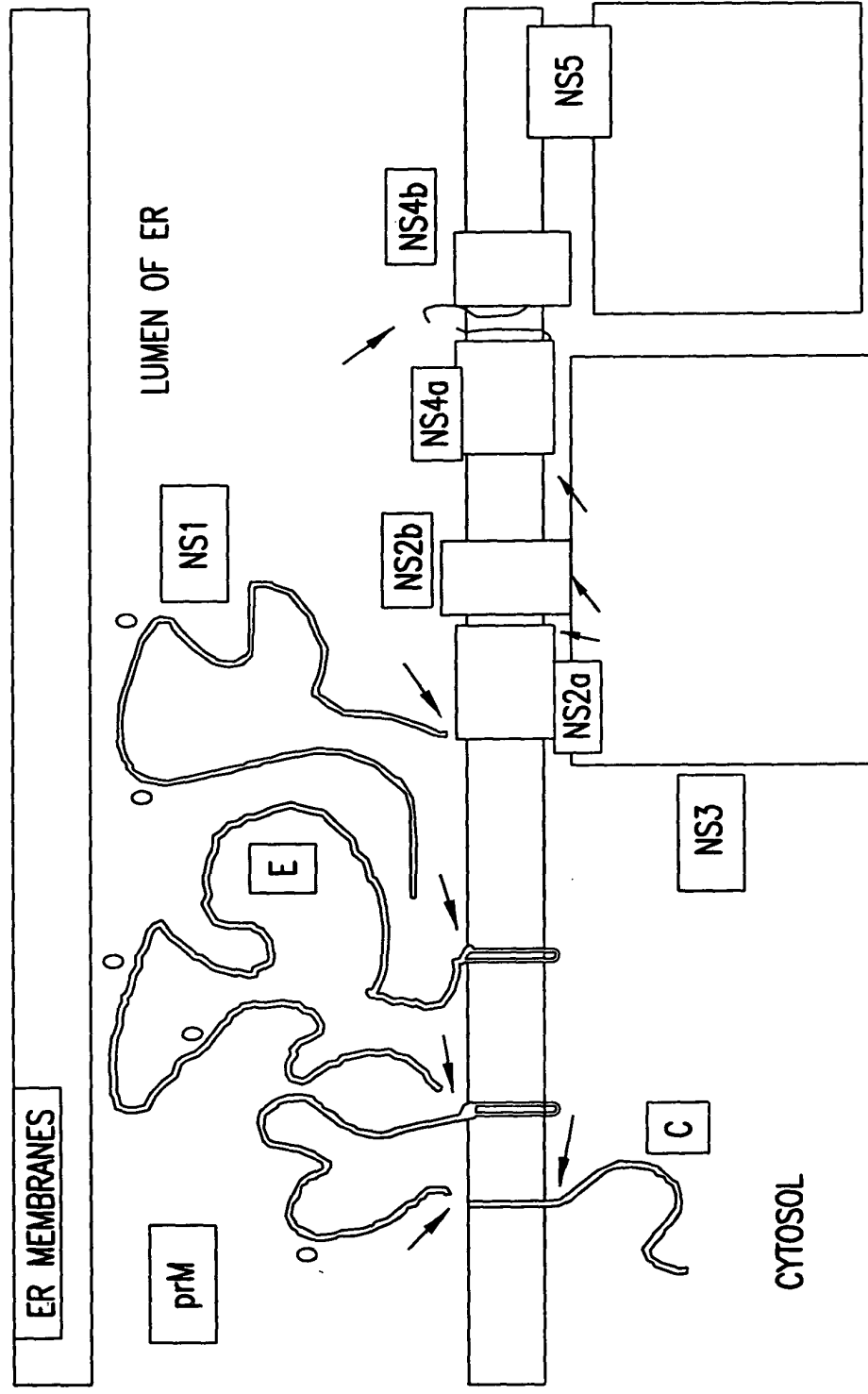


FIG.1A

SCHEMATIC REPRESENTATION OF RECOMBINANT prM AND E PROTEINS

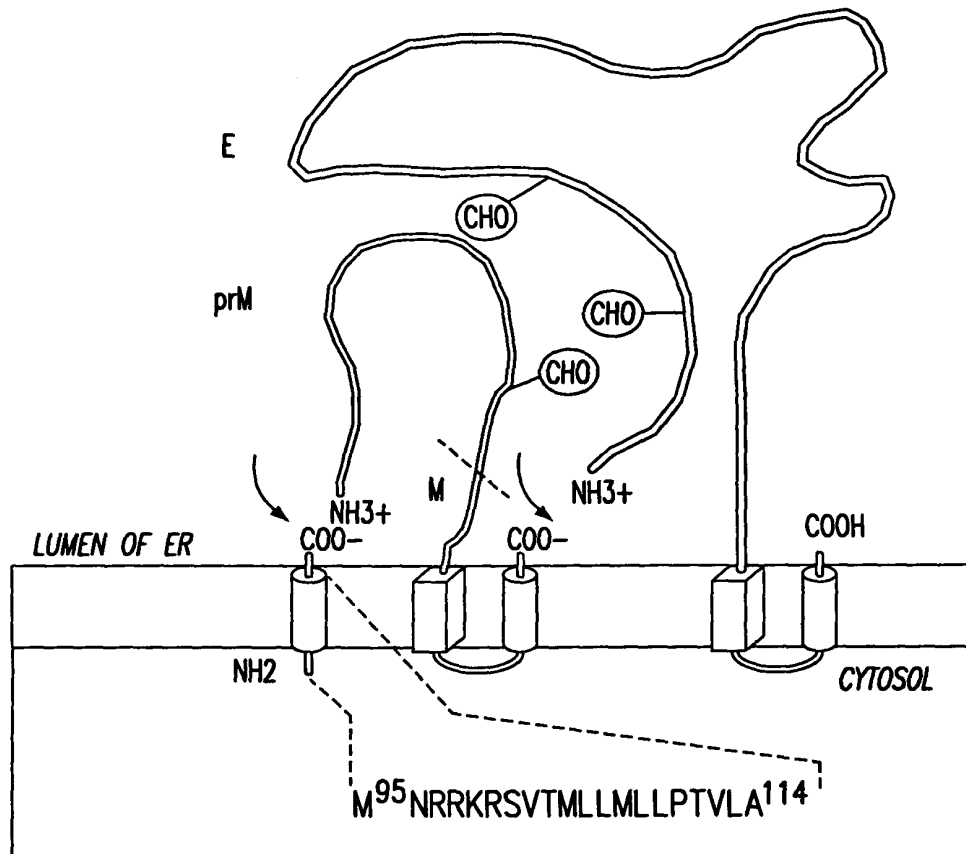


FIG.1B

SCHEMATIC DIAGRAM OF DEN VIRUS PROTEIN E MONOMER

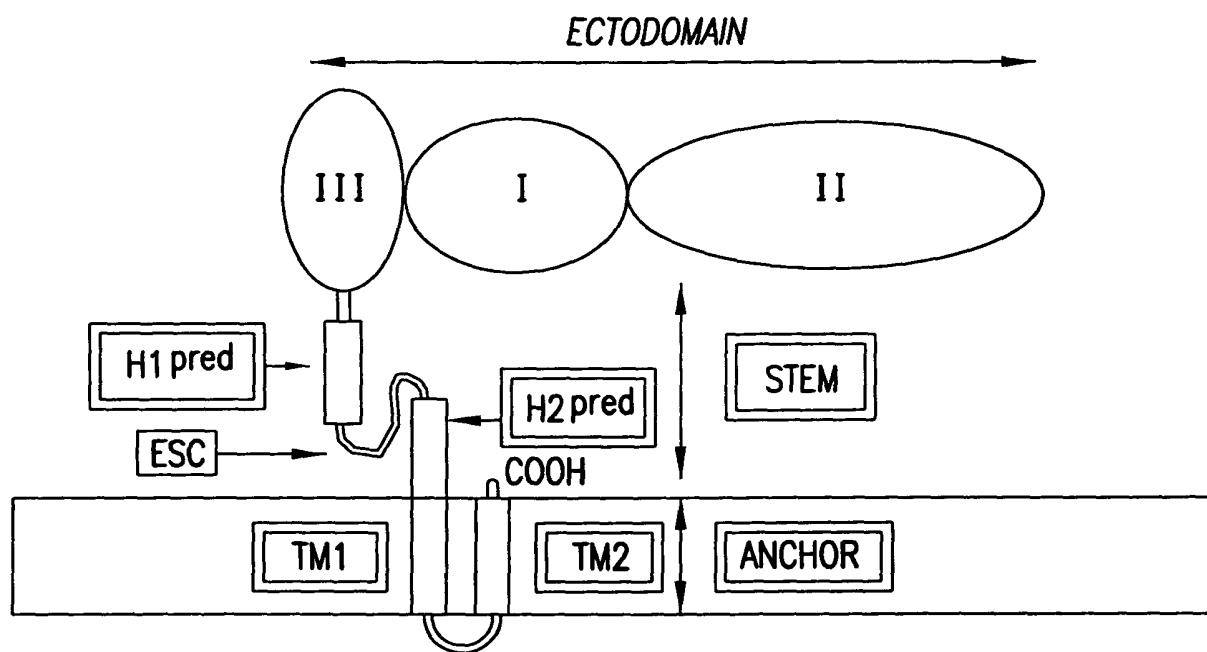
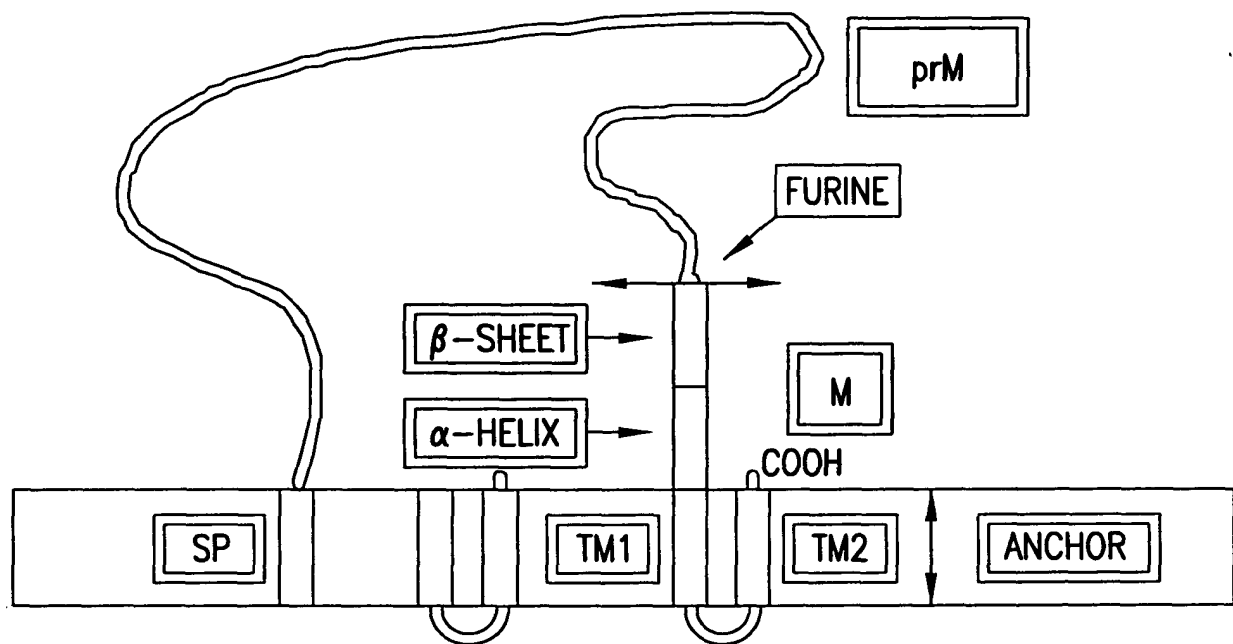


FIG.2A

SCHEMATIC DIAGRAM OF DEN VIRUS PROTEIN prM/M



SCHEMATIC REPRESENTATION OF THE prM PROTEIN STRUCTURE

FIG.2B

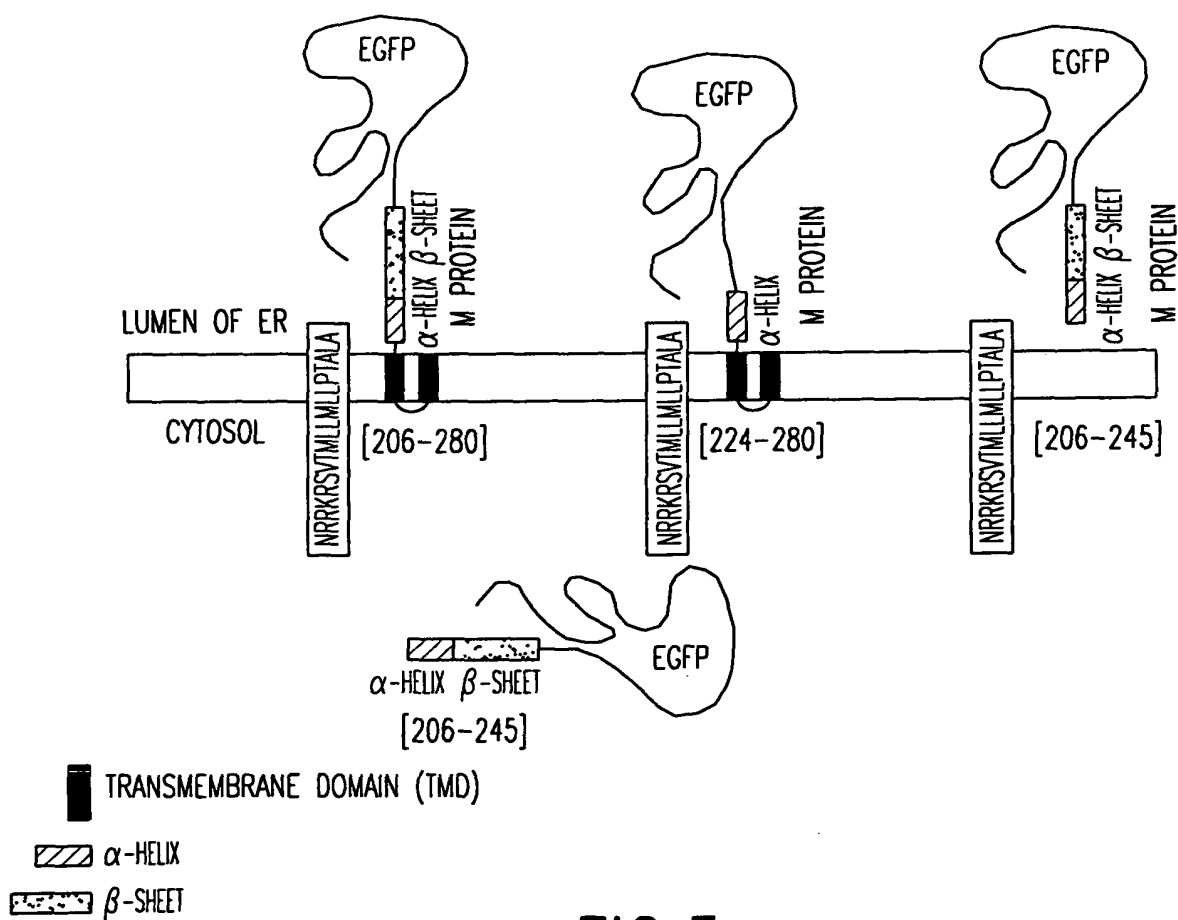
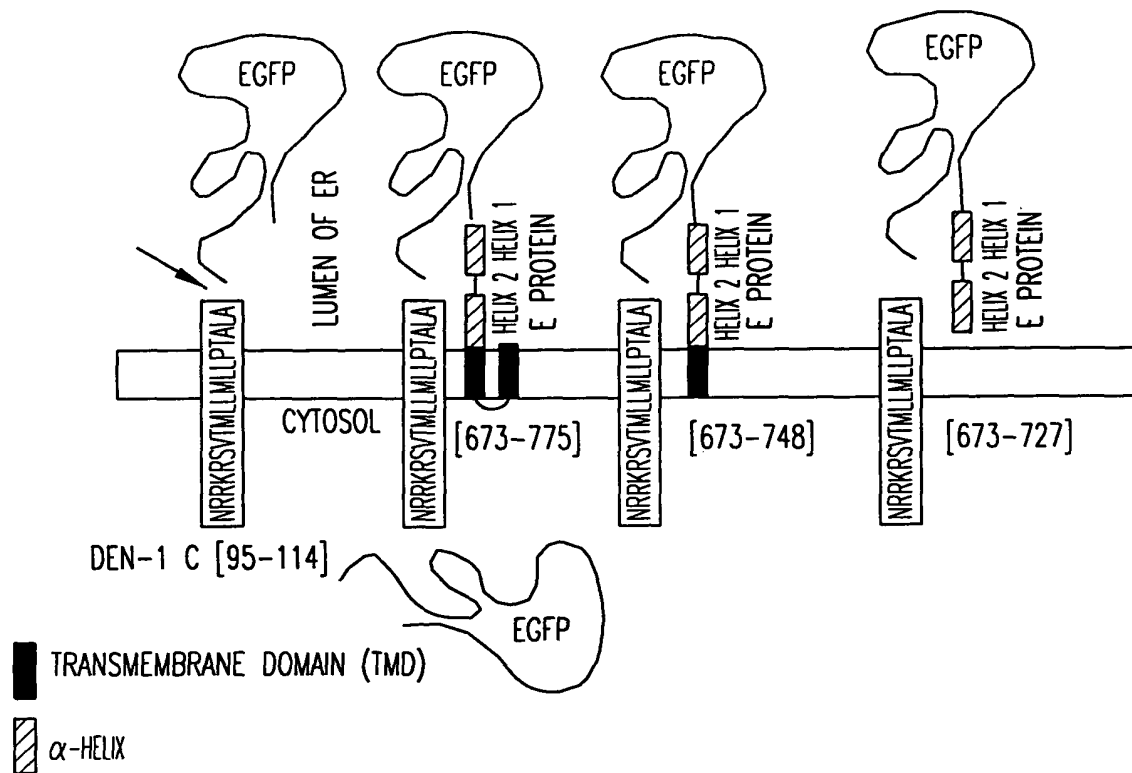


FIG.3

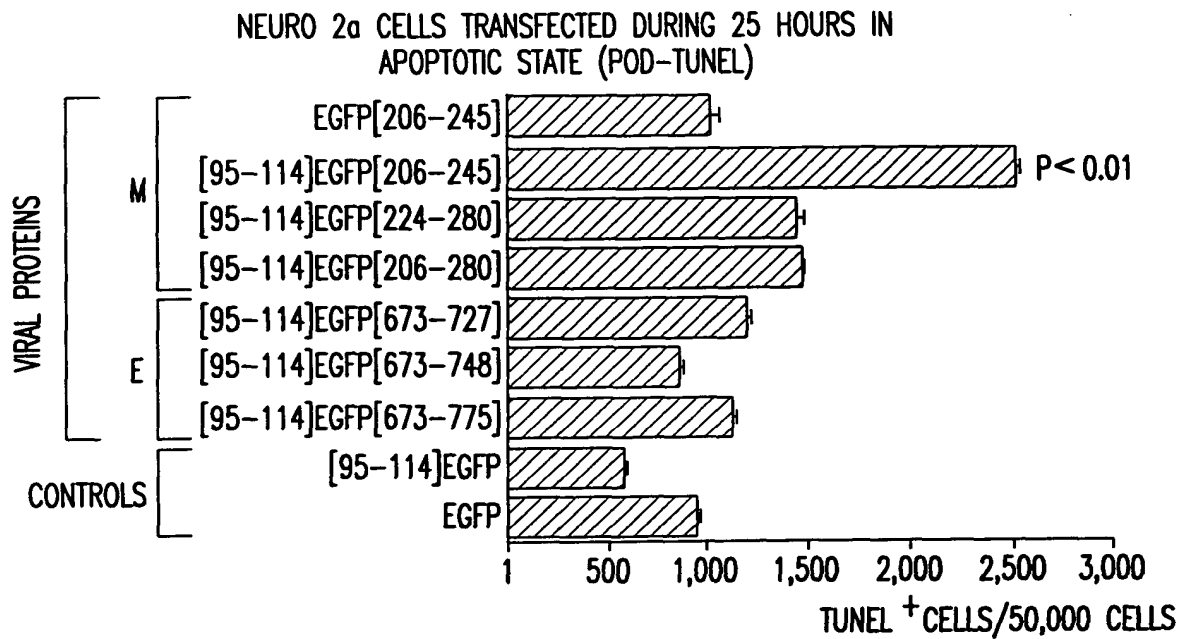


FIG.4A

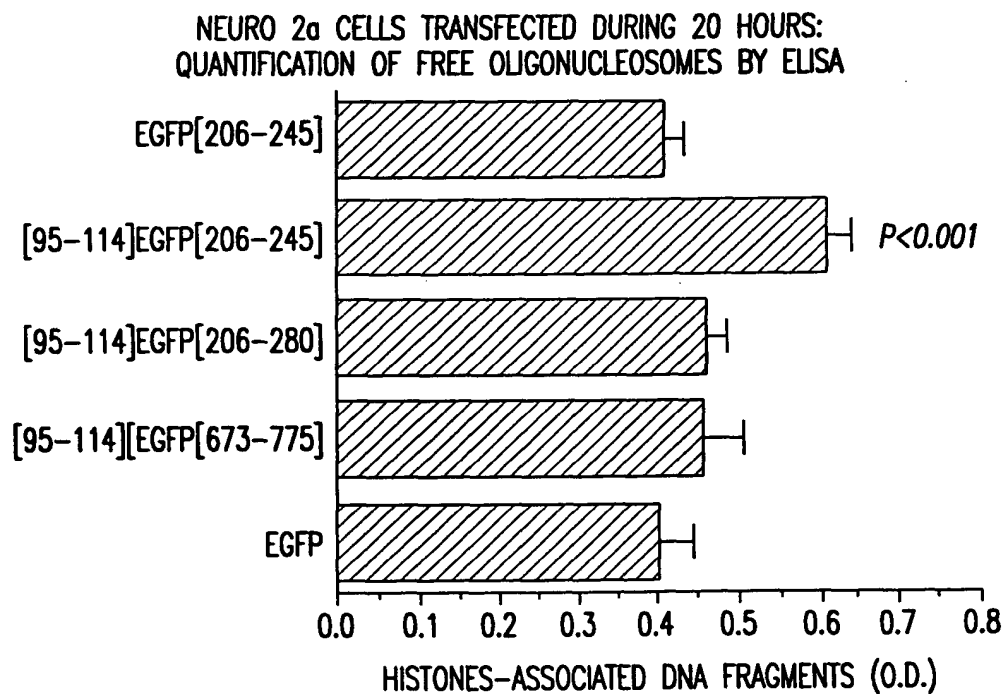


FIG.4B

NEURO 2a CELLS EXPRESSING THE [95-114]EGFP[206-245] FUSION PROTEIN
IN APOPTOTIC STATE (POD-TUNEL)

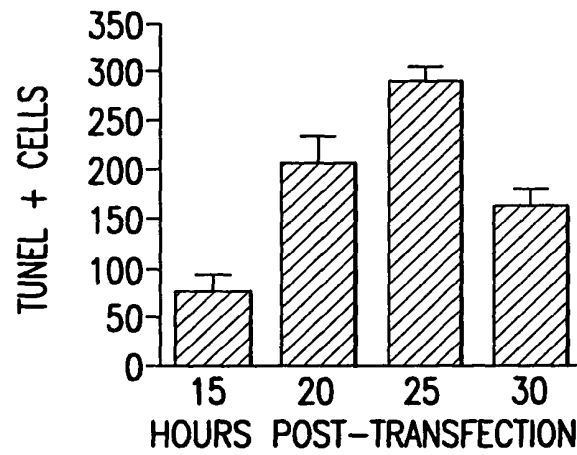


FIG.4C

HepG2 CELLS TRANSFECTED DURING 20 HOURS:
QUANTIFICATION OF FREE OLIGONUCLEOSOMES BY ELISA

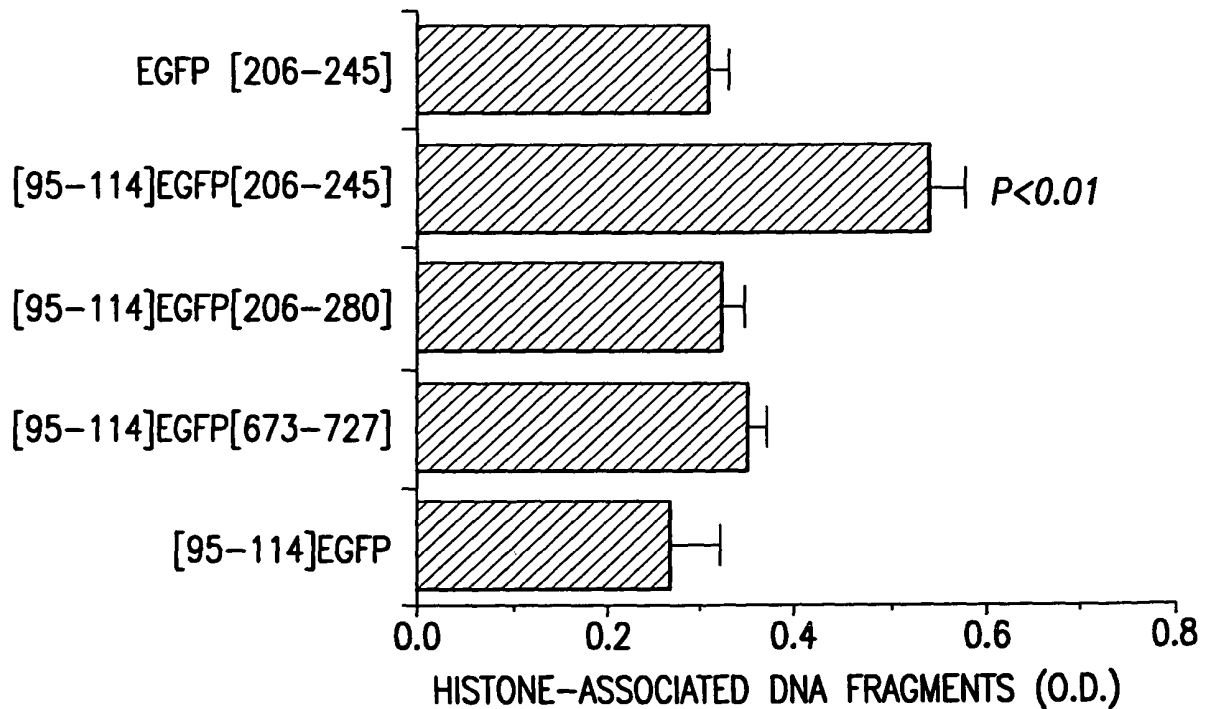


FIG.5A

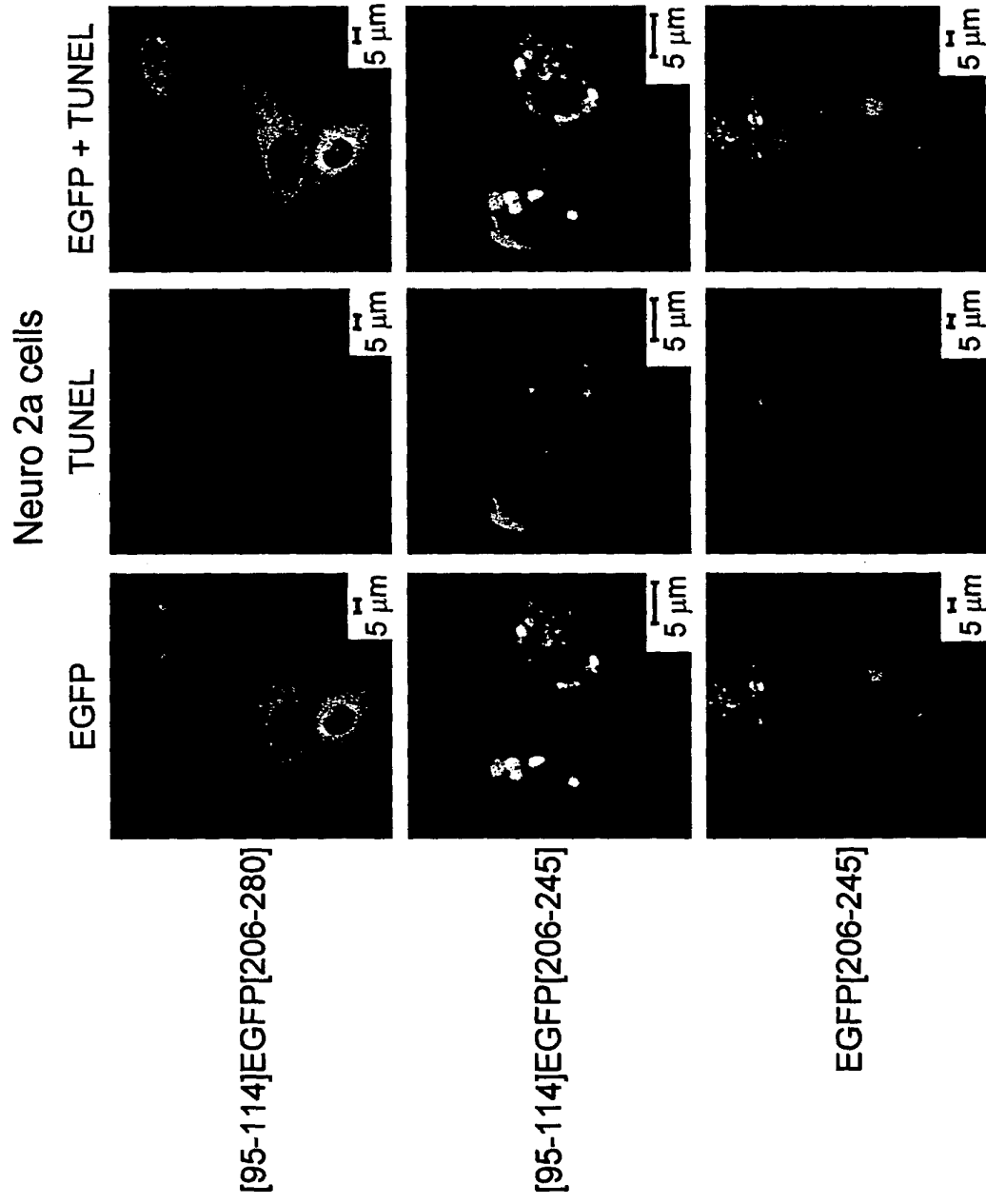


FIG.4D

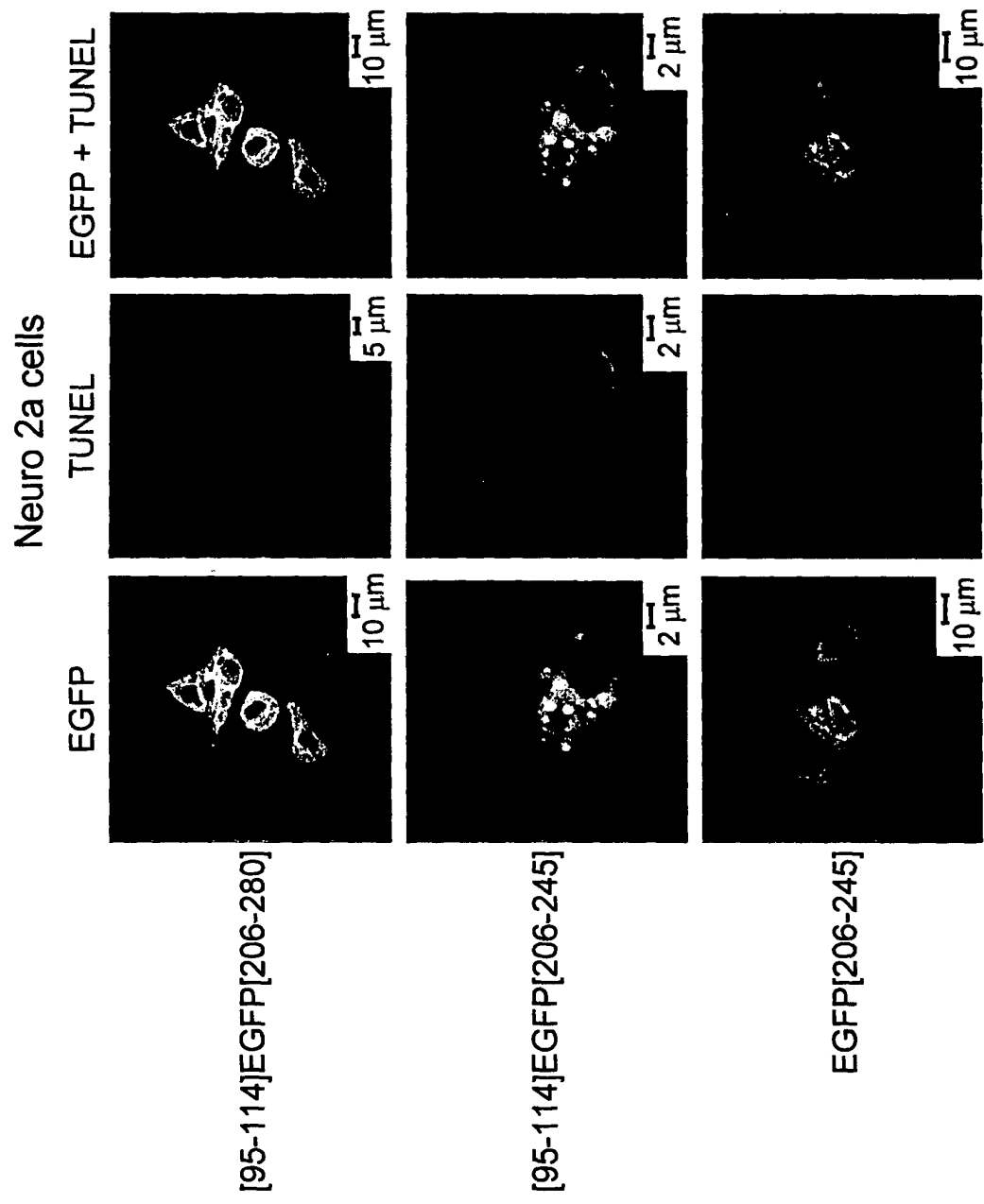


FIG.5B

Sequence of the plasmide p[95-144][EGFP[206-245]]

Sequence in the 5' of the gene EGFP of the plasmide pEGFP-N1 (Clontech, #6085-1, Gene bank accession #U55762)
 591 601 611 621 631 641 651 661 671 EGFP →
 5' G CTA GCG CTA CCG GAC TCA GAT CTC GAG CTC AAG CTT CCA ATT CTG CAG TCG ACG GTA CCG CGG GCG GAT CCA CCG GTC GCC ACC ATG
 Nhe I Sma I

Sequence of the virus Den-1 strain BR/90 in the 5' of the gene of 1'EGFP

sequence DEN-1 →
 M N R R K R S V T M L L M L L P T A L A R E P P V A T M
 5' G CTA ATG AAC AGG AGG AAA AGA TCC GTG ACC ATG CTC CTC ATG CTG CCC ACA GCC CTG GCC CGG GAT CCA CCG GTC GCC ACC ATG
 Nhe I Sma I

Sequence in the 3' of the gene EGFP of the plasmide pEGFP-N1

1380 1390 1400 1410
 L A M • E E L • Y N ochre •
 5' CTC GGC ATG GAC GAG CTG TAC AAG TAA AGC GGC CGC ACT CTA
 BsrG I Not I

Sequence of the virus DEN-1 strain FGA/89 on the 3' of the gene of 1'EGFP

PHVGLGLETRTETWMSSEGAWKQIQKIVETW
 1380 1390
 L A M • E E L • Y S S V A L A I A L R H P opal
 5' CTC GGC ATG GAC GAG CTG TAC AGT TCC GTG GCT CTG GCC GCT TTG AGA CAC CCA TGA TTGGCGCGCGGACTC
 BsrG I Not I

FIG.6

Sequence of the plasmide p[95-114][211-245]

Sequence in the 5' of the gene EGFP of the plasmide Pegfp-N1 (Clontech, #6085-1, Gene bank accession #U55762)

591 601 611 621 631 641 651 661 671 EGFP
 5' G CTA GCG CTA CCG GAC TCA GAT CTC GAG CTC AAG CTT CGA ATT CTG CAG TCG ACG GTA CCG CGG GCC CGG GAT CCA CCG GTC GCC ACC ATG 3'
 Nhe I Sma I

Sequence in the 3' of the gene EGFP of the plasmide pEGFP-N1

1380 1390 1400 1410
 L A M E L • Y N ochre •
 5' CTC GGC ATG GAC GAG CTG TAC AAG TAA AGC GGC CGC ACT CTA 3'
 BsrG I Not I

Sequence of the virus DEN-1 of the plasmide p[95-114][211-245]

sequence [95-114]

591 M N R R K R S V T M L L M L L P T A L A

5' G CTA GCA ATG AAC AGG AGG AAA AGA TCC GTG ACC ATG CTC CTC ATG CTG CTC CCC ACA GCC CTG GCC
 Nhe I

sequence [211-245]

-P H V G L G L E T R T E T W M S S E G A W K Q I Q K V E T W A L R H P

[217-245]

| opal

TGA TTCCGGCGCCGAC 3'

Not I

FIG. 7

Cells expressing the [95-114][211-245] fusion protein

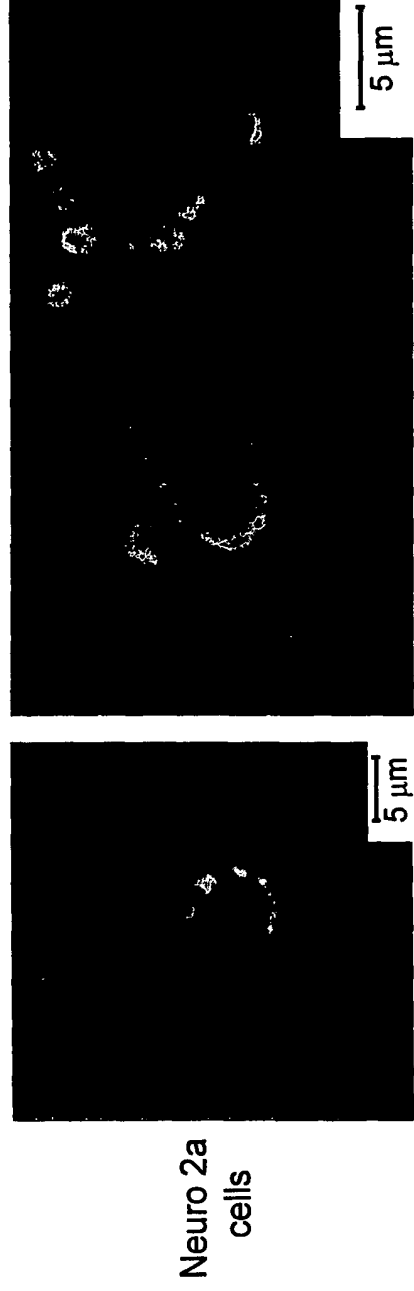
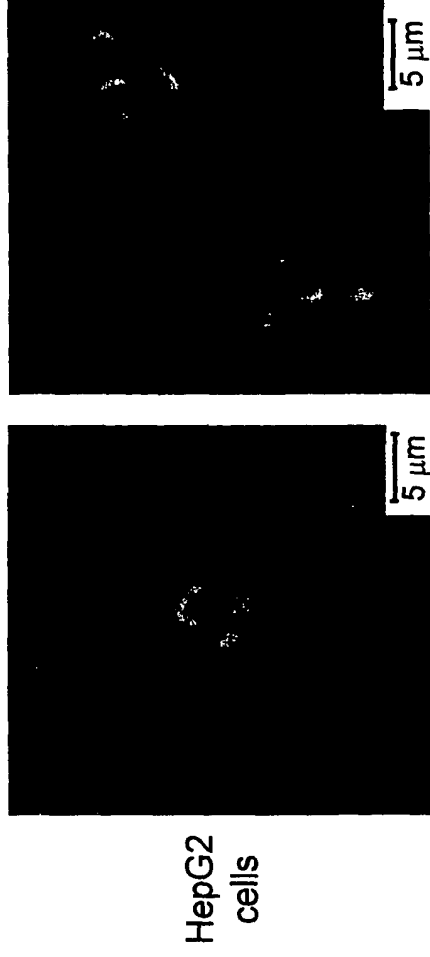


FIG.8

SIMILARITY AND IDENTITY BETWEEN DEN VIRUS AND WN VIRUS

M (West-Nile virus strain IS-98 ST1) 255
215 RSLTVQTHGESTLANKKGAWMDSTKATRYLVKTESWILRNP
SVALAPHVGLGLETRTETWMSSEGAWKQIQKVE TWALRHP 245
206
M (DEN-1 virus strain FGA/89)
32% identity, 66% similarity

Protein M sequences of strains NY99-flamingo382-99 (Genpept access :
AF196835 1) and HNY1999 (Genpept access : AF202541 1) and isolate 2741
(Genpeptnew access : AF206518 1) of West-Nile virus are identical to
that of WN strain IS-98 ST1.

FIG.9

Similarity and Identity

Sequence of the protein M of the virus DEN-1/CD72

M (virus DEN-1, strain FGA/89)

S V A L A P H V G L G L E T R T E T W M S - S E G A W K Q I Q K V E T W A L R H
S P A L A D K A G V G S E Q P T A T W S S V K S S A L R Q I P R C P T V C L Q N
CD72 (aa 56-95) 40% identity

Swiss Prot Library under Accession NosP21855

Peptide 217-242 of the protein M of the virus DEN-1/Box

L E T R T E T W M S S E G A W K Q I Q K DEN-1
L R E R L L G W I Q D Q G G W D G L L S BH2 (Box)
identity 25%, similarity 70%

GenBank Data Library under Accession NosQ 07812

Sequence 217-242 of the protein M from Flavivirus

L E T R T E T W M S S E G A W K Q I Q K	DEN-1
L E T R T E T W M S S E G A W K H A Q R	DEN-2
L D T R T Q T W M S A E G A W R Q V E K	DEN-3
L E T R A E T W M S S E G A W K H A Q R	DEN-4
L S N K K G A W M D S T K A T R Y L V K	KUN
L V N K K E A W L D S T K A T R Y L M K	JE
L V N K K D A W L D S T K A T R Y L T K	MVE
L A N K K G A W M D S T K A T R Y L V K	WN
L A T K N T P W L D T V K T T K Y L T K	SLE
L K T R Q E K W M T G R M G E R Q L Q K	YF
L T G R G H K W L E G D S L R T H L T R	TBE
L T G R G H Q W L E G E A V K A H L T R	LANGAT
M V G T G H A W L K G D N I R D H V T R	POW

DEN-1	GenBank Data Library under Accession Nos	AF 226687
DEN-2	GenBank Data Library under Accession Nos	M 20558
DEN-3	GenBank Data Library under Accession Nos	M 93130
DEN-4	GenBank Data Library under Accession Nos	M 14931
KUN	GenBank Data Library under Accession Nos	D 00246
JE	GenBank Data Library under Accession Nos	U 14163
MVE	GenBank Data Library under Accession Nos	GNWVMV
WN	GenBank Data Library under Accession Nos	AF 196835
SLE	GenBank Data Library under Accession Nos	AF M16614
YF	GenBank Data Library under Accession Nos	K 02747
TBE	GenBank Data Library under Accession Nos	X 07755
LANGAT	GenBank Data Library under Accession Nos	M 73805 et M86650
POW	GenBank Data Library under Accession Nos	L 06436

FIG.10

Sequence of the plasmide p[95-114]EGFP[206-245]DEN-2

Sequence in the 5' of the gene EGFP of the plasmide pEGFP-N1 (Clontech, #6085-1, Gene bank accession #U55762)

591 601 611 621 631 641 651 661 671 EGFP
 • • • • • • • • • →
 5'-G CTA GCG CTA CCG GAC TCA GAT CTC GAG CTC AAG CTT CGA ATT CTG CAG TCG ACG GTA CCG CGG GCC CGG GAT CCA CCG GTC GCC ACC ATG
Nhe I Sma I

Sequence of the virus DEN-1 strain BR/90 in the 5' of the gene of 1'EGFP

sequence DEN-1 →
 M N R R K R S V T M L L M L L P T A L A R E P P V A T M
 5'-G CTA ATG AAC AGG AGG AAA AGA TCC GTG ACC ATG CTC CTC ATG CTG CCC ACA GCC CTG GCC CGG GAT CCA CCG GTC GCC ACC ATG
Nhe I Sma I

Sequence in the 3' of the gene EGFP of the plasmide pEGFP-N1

1380 1390 1400 1410
 L A M• E E L• Y N ochre •
 5'-CTC GGC ATG GAC GAG CTG TAC AAG TAA AGC GGC CGC ACT CTA
BsrG I Not I

Sequence of the virus DEN-2 strain JAM in the 3' of the gene of 1'EGFP

PHVGMGLETRTETWMSSEGAWKIIIVQRIETW
 1380 1390
 L A M• E E L Y R S V A L V! I L R H P opal
 5'-CTC GGC ATG GAC GAG CTG TAC ACA TCA GTG GCA CTC GTT.....ATC TTG ACA CAT CCA TGA GCGGCGCGGACTC
BsrG I Not I

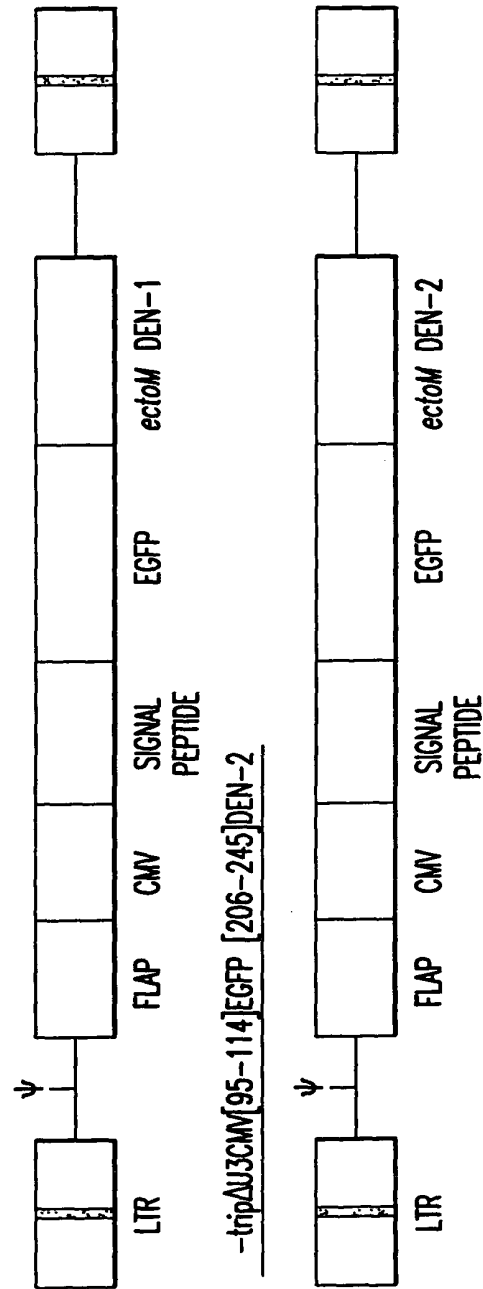
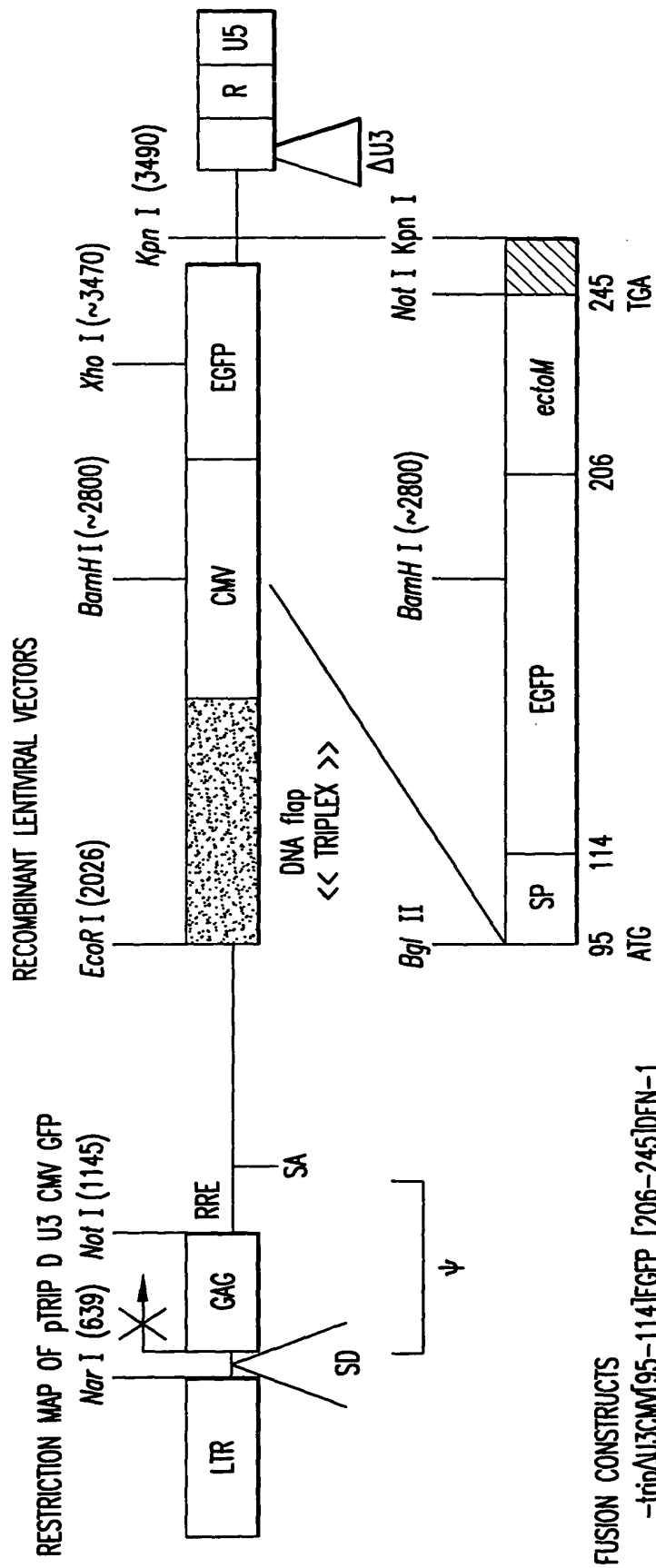


FIG.13

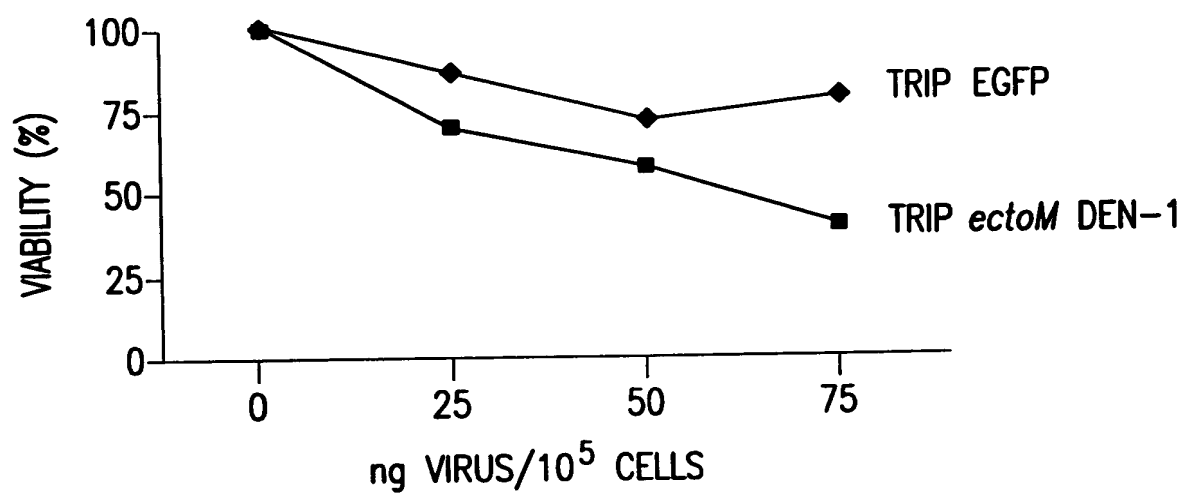


FIG.14

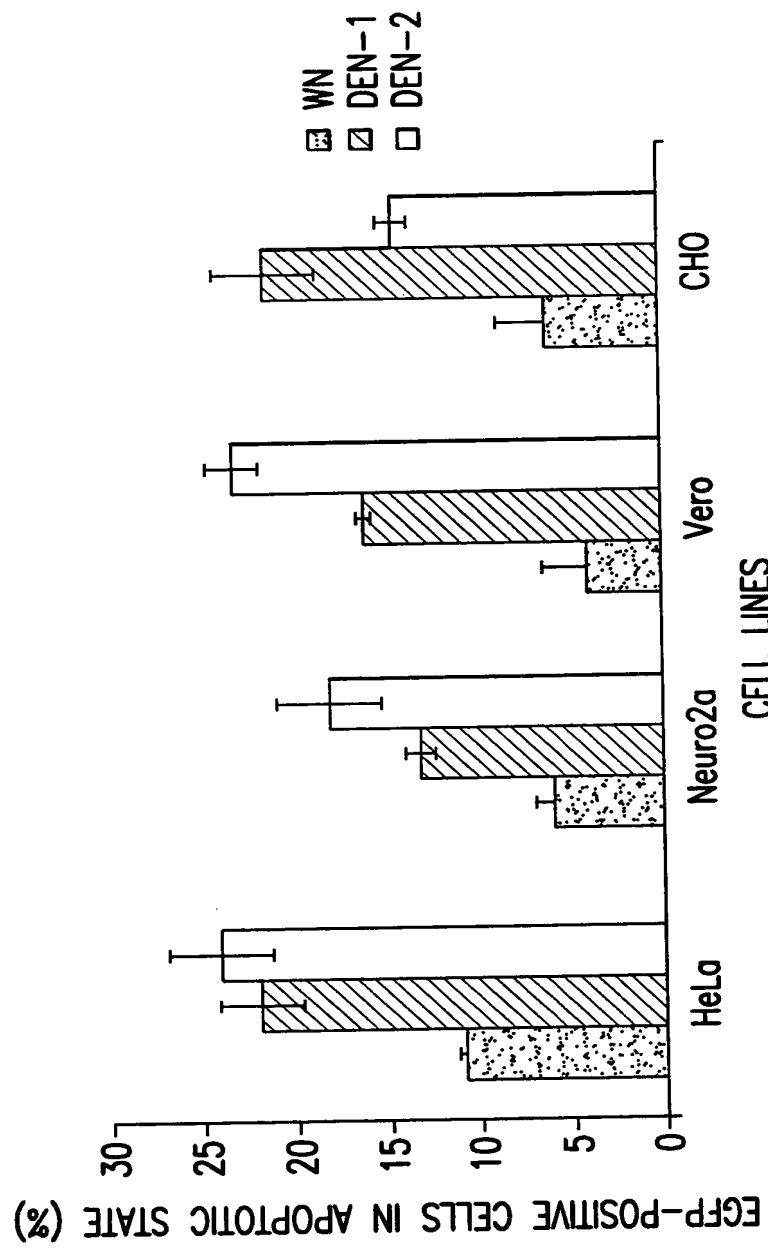


FIG.15